ORIGINAL ARTICLE

# Comparison of Diagnostic Yield in Single Vs Multiple Needle Passes in Endoscopic Ultrasound Guided Fine Needle aspiration cytology in Abdominal Solid Lesions and Abdominal lymphnodes at Tertiary Care Centre

**Shivani Meena*, Dilip Ramrakhiani**, Rimjhim Shrimal***, Sandeep Nijhawan****, Ashok Jhajharia*****, Deepti Yadav***

## ABSTRACT

**Introduction:** Endoscopic ultrasound-guided fine needle aspiration (EUS-FNA) is the accurate diagnostic method for abdominal solid masses, mediastinal masses and lymph nodes. Endoscopic ultrasound has proven to be a highly sensitive tool for diagnostic lesions in and adjacent to the gastrointestinal tract. EUS-FNA is not a difficult technique but it requires adequate experience.The present study was done to compare the diagnostic yield of single needle pass vs multiple needle passes.

**Methods**: It was an observational hospital based study. There were two groups made with 30 samples each. Duration of study was 6 months. Consecutive sample were taken with inclusion criteria of age more than 15 years and solid masses. Fine needle aspiration by done using endoscopic ultrasound in department of gastroenterology.

**Results:** Two groups were made which were single needle and multiple needle passes. Mean number of passes in pancreatic masses and lymph node were 3.1 and 2 respectively. Cellularity was significantly different in the two groups having higher cellularity in group II. There was no significant difference in the definitive diagnosis between the two groups.

**Conclusion:** Higher cellularity observed in group of patients with multiple needle passes was statistically significant but the higher diagnostic yield observed in this group was not statistically significant due to limited sample size.

**Keywords:** Endoscopic Ultrasound, FNAC, Needle passes, Diagnostic Yield

## INTRODUCTION

Endoscopic ultrasound-guided fine needle aspiration (EUS-FNA) is the accurate diagnostic method for abdominal solid masses and mediastinal masses and lymph node and its accuracy is affected by various FNA methods and EUS equipment.

Endoscopic ultrasound (EUS)-guided fine needle aspiration (FNA) has been used for diagnosis of pancreatic solid masses. EUS-FNA has a reported sensitivity of 54% to 95%, a specificity of 71% to 100%, and an accuracy of 85% to 90%. The foundation for the diagnostic accuracy of EUS-FNA is obtaining adequate tissue, and it could be influenced by several variables, including the size of the lesion, location of the lesion, needle gauge, needle type, use of a stylet and suction, number of needle passes, the endosonographer's skill and experience, and on-site cytopathology evaluation.

Endoscopic ultrasound has proven to be a highly sensitive tool for diagnostic lesions in and adjacent to the gastrointestinal tract. EUS-FNA is not a difficult technique but it requires adequate experience. Interestingly, some of the easiest cases provide information that can have a tremendous impact on patient management[1,2].

The goal of performing FNA is to obtain a positive diagnosis in the quickest possible time with least number of passes. The number of passes to be made depends on the presence or absence of on-site cytopathologist for assessment of specimen adequacy, establishment of onsite diagnosis and to guide the need for further sampling. In the absence of an on-site pathologist,

*PG Resident, Department of Pathology, SMS Medical College, Jaipur.
**Senior Professor, Department of Pathology, SMS Medical College, Jaipur.
***Senior Demonstrator, Department of Pathology, SMS Medical College, Jaipur.
****Senior Professor, Department of Gastroenterology, SMS Medical College, Jaipur.
*****Assistant Professor, Department of Gastroenterology, SMS Medical College, Jaipur.
**Corresponding Author:**
Dr. Dilip Ramrakhiani
Department of Pathology, SMS Medical College, Jaipur.
**Email :** dilip.ramrakhiani@gmail.com

adequate passes should be performed to avoid the need for repeat procedure[3].

EUS-FNAC help us to get diagnostic material which give us information to guide disease specific therapeutic intervention. This techniques causes prevention of unnecessary operative procedure. The usefulness of EUS-FNA depends on several factors. In addition to the experience of the endoscopist, adequate sampling, adequate sample processing, better communication between the cytopathologist and the endoscopist, accurate interpretation by the cytopathologist, and the ability to determine the need for additional samples required for ancillary studies are needed for effective diagnosis[4].

Several studies in the last year focused on the technical aspects of EUS-FNA like optimal needle choices, variety of sampling method and different techniques of specimen[5,6]. Although there is wide use of EUS-FNA, there still exists a wide variation in the number of samples required to ensure acquisition of diagnostic material from different kind of lesions[7].

In the presence of an onsite pathologist, the smears are quickly processed and examined by light microscopy in the endoscopic suit and immediate feedback is given to endosonographer. This information can be used in guiding the number of EUS-FNA passes required to obtain a final diagnosis. It evaluates whether the aspirate is diagnostic or non-diagnostic[8]. When on-site cytological evaluation is unavailable, ESGE suggests performing of three to four needle passes with an FNA needle or two to three passes with a fine needle biopsy needle[9].

The present study was conceived to assess and compare the diagnostic yield of sample obtained from EUS-FNA from abdominal solid lesions and lymphnode in a single needle pass v/s multiple needle passes.

## MATERIALS AND METHOD

**Study Design**: Descriptive observational study.

**Sample Size:** Sample size was calculated with 95% confidence level and alpha error of 0.05 assuming 57.7% and 85.7% diagnosis was achieved in patients on pass one by standard and fanning technique respectively as given by Bang et al[10]. At a study power of 80% the required sample size was 30 patients for each group.

**Study Duration**: Duration of study was July 2019 to Jan 2020.

**Study Universe:** EUS Guided FNA done in one needle pass and multiple needles pass in Gastroenterology Department of Medical College and attached hospitals, Jaipur.

**Inclusion Criteria:** The inclusion criteria were as follows:

1. Age $\geq$15 years,
2. Abdominal solid masses including lymph nodes identified by the investigational modalities.

**Exclusion Criteria**: The exclusion criteria were as follows:

1. Coagulopathy (international normalized ratio of >1.5 or platelet count of <50,000/mm3)
2. Presence of intervening blood vessels, and altered gastrointestinal anatomy.
3. Cystic Masses.

**Sampling Technique**: Every consecutive eligible EUS guided FNA done in Gastroenterology Department at SMS Hospital, Jaipur.

**Study Tool**: Fine Needle Aspiration (FNA) done by endoscopic ultrasound in Gastroenterology Department in the patient who had G.I. solid lesions and/or lymph nodes.

### EUS-FNA Procedures

EUS-FNA procedures were performed using a standardized method in patients who were under conscious sedation with intravenous midazolam and propofol. All procedures were carried out using a linear array echo endoscope (GF UCT180; Olympus Medical Systems, Tokyo, Japan) in conjunction with EVIS EXTRA CLV-180 light source.

The needle size was chosen to fit the situation randomly by endosonographer. A standard 19-, 22-, or 25-G FNA device (Echo Tip; Cook Medical, Bloomington, IN) was employed for EUS-FNA. The capillary (slow pull) technique was employed for EUS-FNA mostly. In some cases, we applied suction technique during EUS-FNA in order to increase the quantity of the FNA sample. Pancreatic head masses were approached from the duodenum, whereas pancreatic body and tail masses were

accessed from the stomach. The adequacy of obtained specimens was judged by the presence of macroscopic material without cytopathologist, and the puncture was repeated until adequate specimens are obtained. After the masses were punctured by the needle, the stylet was withdrawn and the needle moved backward and forward within the masses 10 to 15 times per pass. The needle was then removed. The aspirated specimen was expressed onto slides by reinsertion of the stylet within the needle and air flushing, if needed.

**Statistical Analysis:** Sample size was selected on the basis of sample size calculation formula for difference of effect size proportions in the two groups. The statistical tests used in the study are unpaired Student's *t*-test for continuous variables and Chi-square test for categorical variables. Mann-Whitney test was used to assess the diagnostic yield by different number of needle passes in EUS-FNA. SPSS version 20.0 was used for statistical analysis. Data were expressed as mean ± standard deviation. Value of $P < 0.05$ is considered significant.

## RESULTS

Patient in group I were slightly younger than group II with mean age of 47.4 years in group I as compared to group II (49.6years)(Table 1). In the both groups males were predominant with overall affected male to female ratio of 3:1.Including both groups (single pass and multiple passes) pancreas was the most common site (28.5%) followed by abdominal lymph nodes (26.6%). (Table 2)

**Table 1: Baseline characteristics of EUS-FNA according to number of the needle passes used in abdominal organs and lymph nodes**

| Variables | Single needle pass (Group I) | Multiple needle passes (Group II) |
|---|---|---|
| Age in years (range) | 47.4±15.36 (15-75) | 49.6±15.36 (30-70) |
| Sex (Males:Females) | 22:8 | 23:7 |
| Size of Tumour | 24.7±13.3 (10-70) | 29.97±19.37 (10-80) |
| Cellularity | 18/30 | 26/30 |
| Definitive Diagnosis | 25/30 | 27/30 |

**Table 2: Distribution of patients according to target regions**

| EUS-FNA Site | No. of Cases (60) | % of Cases (Out of 100) |
|---|---|---|
| Pancreas | 17 | 28.5% |
| Abdominal Lymph Node | 16 | 26.6% |
| Mediastinal Lymph Node | 9 | 15% |
| Liver | 8 | 13.5% |
| Common Bile Duct | 4 | 6.6% |
| Spleen | 3 | 5% |
| Duodenum | 1 | 1.6% |
| Gastric Mass | 1 | 1.6% |
| Gall Bladder | 1 | 1.6% |

In second group of multiple passes more no. of passes were three (43.3%) followed by two passes (26.6%) and 4 passes (16.6%) (Figure 1). Mean number of needle passes used in pancreas were 3.1 needle passes for diagnosis. While in 25 cases of lymph nodes mean number of needle passes were 2.

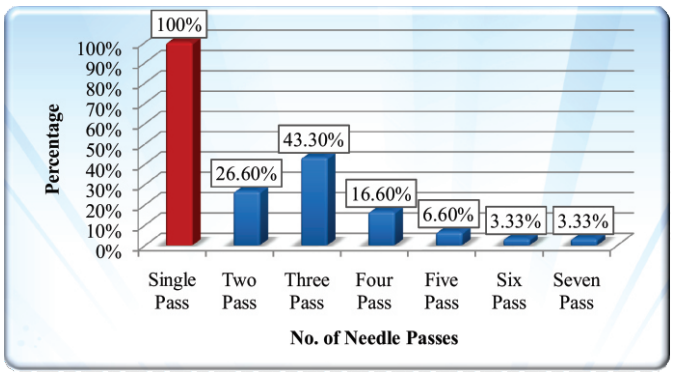


**Figure 1: Distribution of patients according to no. of needle passes**

In 30 cases where single needle was passed using EUS guided FNA, there was cellularity in 22 cases (73.3%) and 8 cases were non cellular (26.6%). In EUS-FNA using multiple passes (⩾ 2 passes), the frequent number of passes used were 3 in which cellularity was found in 8 cases and 1 case was non cellular. Cellularity was 26/30 in Group II and it was significantly different from Group I (p=0.0013).

**Table 3: Distribution of patients according to number of pass and Cellularity (Group II)**

| No. of the Needle Passes | Cellularity No. of Cases) | Non-Cellular (No. of Cases) |
|---|---|---|
| 2 Passes | 7 | 2 |
| 3 Passes | 8 | 1 |
| 4 Passes | 6 | 1 |
| 5 Passes | 2 | 0 |
| 6 Passes | 1 | 0 |
| 7 Passes | 1 | 0 |

The final cytopathological diagnosis was reported (malignant or benign cases) in14 cases (46.6%) out of 30 cases in Group I as compared with 15 cases out of 30 cases (50%) in group II. Other categories included were inconclusive diagnosis and non-diagnostic cases (Table 4).

**Table 4: Distribution of cases according to number of needle passes and cytopathological diagnosis**

| | Diagnosis | Group I (Single Pass) | Group II (Multiple Pass) |
|---|---|---|---|
| Malignant cases | Pancreatic Carcinoma | 4 (13.3%) | 4 (13.3%) |
| | Lymphoma | 3 (10%) | 1 (3.4%) |
| | Hepatocellular Carcinoma | 2 (6.6%) | 4 (13.3%) |
| | Metastatic lymph node | 4 (13.3%) | 4 (13.3%) |
| Benign cases | GIST | 1 (3.4%) | 0 |
| | Granulomatous lesion | 1 (3.4%) | 0 |
| | Psuedocyst of pancreas | 0 | 1 (3.4%) |
| | Tubercular lymphadenopathy | 4 (13.3%) | 3 (10.1%) |
| | Reactive hyperplasia | 4 (13.3%) | 4 (13.3%) |
| | Inconclusive Diagnosis | 2 (6.7%) | 5 (16.6%) |
| | Non Diagnostic Cases | 5 (16.6%) | 4 (13.3%) |

On statistical analysis, there was no significant difference in the definitive diagnosis between single pass and multiple pass needle (Table 5).

**Table 5: Association between number of passes and cellularity**

| | Single pass n (%) | Multiple pass n(%) | Total | p-value |
|---|---|---|---|---|
| Cellularity present | 18 (40.9) | 26 (59.1) | 44 (100) | 0.013 |
| Non-cellular | 12 (75) | 4 (25) | 16(100) | |
| Total | 30(50) | 30(50) | 60 (100) | |

*Fisher exact test

## PHOTOGRAPHS

### Group I (Single needle pass EUS FNA)

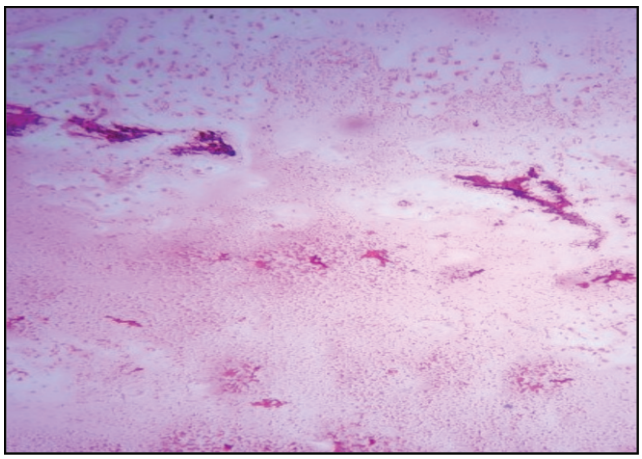

**Figure 2: EUS FNA from Gall Bladder Mass- hypocellular smear showing few cells enmeshed in blood clot. (H&E, 40X)**

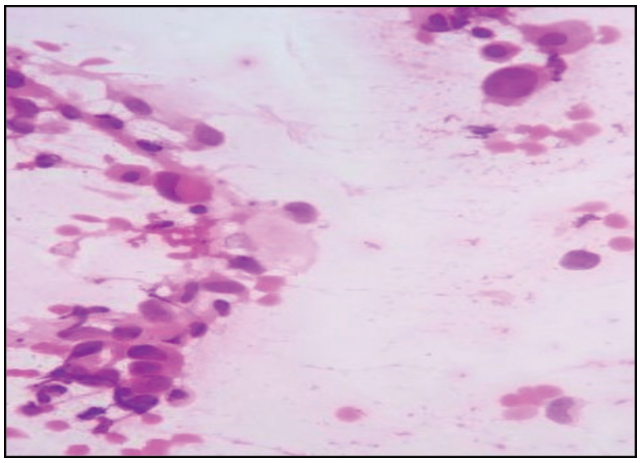

**Figure 3: High power view showing few benign cells only. Inadequate for diagnosis (H&E, 400x)**

**GROUP II (MULTIPLE NEEDLE PASSES EUS-FNA)**

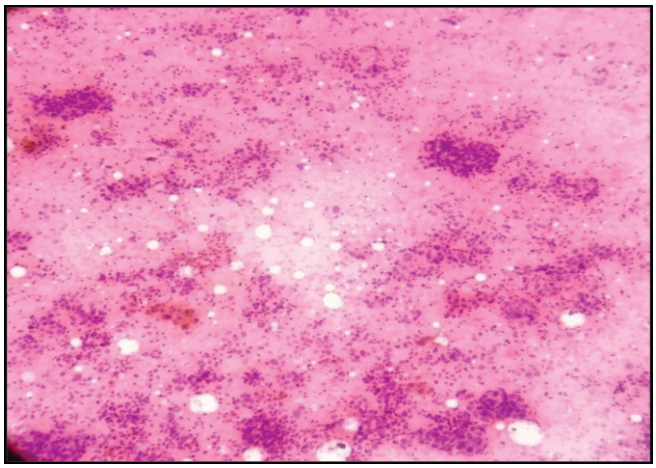

**Figure 4: EUS FNA from head of pancreas- abundant cellularity (H&E, 40X)**

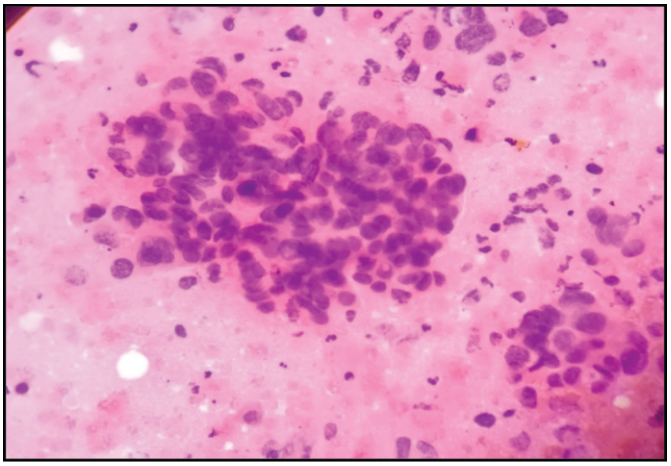

**Figure 5: High power view showing cells in adenomatous pattern with pleomorphism and high N:C ratio- Moderately differentiated adenocarcinoma of pancreas (H&E 400X)**

## DISCUSSION

In present study there were two groups including single needle pass and multiple needle passes in EUS – FNA cases. The number of needle passes and diagnostic yield were not significantly affected with the age of the patients. As Iglesias et al. conducted a studied 182 cases and found no association with age, sex, location and size of lesions[11]. In the present study, more common target sites for EUS-FNA were pancreas (17 out of 60 cases) and lymph nodes (16 out of 60 cases). Similarly, Bang et al conducted a prospective study of 95 consecutive patients who underwent EUS FNA, for diagnosis or staging, of 95 sites: pancreas (33), Lymph node (43), and miscellaneous (19)[10].

In group II (multiple number of needle passes) frequently used number of needle passes were three (13 out of 30 cases) with mean number of needle passes in current study as 3.26. Itoi et al found that even without ROSE, mean 2.88 needle passes were adequate for diagnosis with 93.3% accuracy[12]. When ROSE is not available there is more number of needle passes needed. Which is in concordance in our findings in multiple needle passes. There were 30 cases of single needle passes, diagnosis was possible in 22 cases out of 33 (cellularity 73.3%). Lim et al described that single pass can also be used for definitive diagnosis which varies according to target sites[13].

There was more cellularity in cases of group II. 26 cases out of 30 cases (86%) were cellular for diagnosis and p-value for this association was 0.013 which is significant. Cellularities were affected by number of needle passes. For pancreas and lymph nodes the mean number of passes required were 3.1 and 2 respectively. In our study greater cellularity yield was observed in multiple needle passes as compared to single pass (statistically significant, p-value=0.013). This is in concordance with the various studies. Erickson et al. showed that 5 to 6 passes were required for pancreatic masses, meant that multiple passes were needed to obtain accurate diagnosis[14]. Also, Le Blanc et al. illustrated in a study that increase in sensitivity from 17% to 87% when more than 7 passes were made with a 22 G needle into pancreatic masses[15].

Turner et al. demonstrated in their study that 3 to 4 passes were necessary to achieve a diagnostic accuracy of 80%[16]. Suzuki et al. described in a study involving 25 G needle, 4 passes were found to be sufficient for EUS FNA of solid pancreatic lesions[17]. Bluen et al. stated in an article that several factors can be responsible for diminished accuracy of abdominal and mediastinal area. More number of needle passes need to make out diagnosis[18].

Per-pass analyses of data from some studies in patients with pancreatic masses showed that three to four passes with an FNA needle to achieve high rates of diagnostic samples and high sensitivity for malignancy, which is more than 90%[19-24]. A lower number of passes was associated with suboptimal performance. On the other hand, increasing the number of needle passes more than four (FNA) or three marginally improved the results[20,23,24]. However, also for the smaller tumors, increasing the number of passes beyond four only marginally improved the sensitivity. Per-pass analysis in patients with lymphadenopathy found that sensitivity for malignancy reaches 100% after three passes with an FNA needle[24]. Binmoeller attributed that cellularity and adequacy differ when there is on site cytopathologist was not available[25]. Even ESGE criteria recommend multiple passes for various organs as was observed in our study[26].

The diagnostic yield in the multiple pass group was 51% and this was higher than the diagnostic yield in single needle pass group (49%). Although this result is not statistically significant due to limited sample size (limitation of study) but this has also been shown in several studies mentioned previously involving larger number of patients that multiple needle passes associated with high diagnostic adequacy.

## CONCLUSION

In this study, greater cellularity and diagnostic yield were observed in group of patients subjected to multiple needle passes. The higher cellularity observed in group of patients with multiple needle passes was statistically significant but the higher diagnostic yield observed in this group was not statistically significant due to limited sample size.

## REFERENCES


1. O'Toole D, Palazzo L, Arotçarena R, Dancour A, Aubert A, Hammel P, et al,. Assessment of complications of EUS-guided fine-needle aspiration. Gastrointest Endosc, 2001: 53 (4): 470–4.
2. Klapman JB, Logrono R, Dye CE, Waxman I. Clinical Impact of on-site cytopathology interpretation on endoscopic ultrasound – guided fine needle aspiration. Am J Gastroenterol. 2003; 98(6):1289-94.
3. Vilmann P, Saftoiu A. Endoscopic ultrasound guided fine needle aspiration biopsy:equipment and technique. J Gastroenterol Hepatol. 2006; 21(11):1646-55.
4. Bhutani MS, Logrono R, Manoop S. Endoscopic ultrasound-guided fine-needle aspiration cytology for diagnosis above and below the diaphragm. J Clin Ultrasound; 2005; 33(8):401-11.
5. Tharian B, Tsiopoulos F, George N, Pietro SD, Attili F, Larghi A, et al,. Endoscopic ultrasound-guided fine needle aspiration technique and application in clinical practice. World J GastrointestEndosc. 2012: 4(12):532–44.
6. Lachter J, Rubin A, Shiller M, Lavy A, Yasin K, SuissaA,et al,. Linear EUS for bile duct stones. GastrointestEndosc;2000:51(1):51–54.
7. Lachter J, Zelikovsky S.EUS changes diagnosis from pancreatic to gallbladder carcinoma. InternJGastrointest Cancer. 2002;32(2-3);161–4.
8. Hirdes MMC, Vleggaar FP.Performance of EUS-FNA for mediastinal lymphadenopathy: impact on patient management and costs in low-volume EUS centers. SurgEndosco. 2010; 25(9), 3124-5.
9. Petrone MC, Arcidiacono PG. Basic technique in endoscopic ultrasound-guided fine needle aspiration for solid lesions: How many passes? Endosc Ultrasound. 2014 ;3(1);22-7
10. Bang JY, Ramesh J, Trevino J, Eloubeidi MA,Varadarajulu S. Objective assessment of an algorithmic approach to EUS-guided FNA and interventions. GastrointestEndosc. 2013;77 (5):739–44.
11. Iglesias-Garcia J, Dominguez-Munoz E, Lozano-Leon A, Abdulkader I, Larino-Noia J, Antunez J, et al,. Impact of endoscopic ultrasound-guided fine needle biopsy for diagnosis of pancreatic masses. World J Gastroenterol. 2007;13(2);289-93
12. Itoi T, Tsuchiya T, Itokawa F, Sofuni A, Kurihara T, Tsuji S, et al,. Histological diagnosis by EUS-guided fine-needle aspiration biopsy in pancreatic solid masses without on-site cytopathologist: a single-center experience. Dig Endosc. 2011;23 Suppl 1:34-8.
13. Lim LG, Lakhtakia S, Ang TL, Vu CK, Dy F, Chong VU, et al,. Factors determining diagnostic yield of endoscopic ultrasound guided fine-needle aspiration for pancreatic cystic lesions: A multicentre Asian study. Dig Dis Sci. 2013:58(6);1751-7
14. Erickson RA, Sayage Rabie L, Beissner RS. Factors predicting the number ofEUS guided fine needle passes for diagnosis of pancreatic malignancies. GastrointestEndosc. 2000;51(2):184-90.
15. LeBlanc JK, Ciaccia D,Al-Assi MT, Mc Granth K, Imperiale T, Tao LC, et al,. Optimal number of EUS-guided fine needle passes needed to obtain a correct diagnosis.GastrointestEndosc. 2004;59 (4); 475–81.
16. Turner BG, Cizginer S, Agarwal D, Yang J, Pitman MB, Brugge WR, et al,. Diagnosis of pancreaticneoplasia with EUS and FNA; A report of accuracy.GastrointestEndosc. 2010;71(1);91-8
17. Suzuki R, Irisawa A, Bhutani MS, Hikichi T, Takagi T, Sato A, et al,. Prospective evaluation of the optimal number of 25 gauge needle passes for endoscopic ultrasound guided fine needle aspiration biopsy of solid pancreatic lesions in the absence of an onsite cytopathologist. Dig Endosc. 2012;24(6);452-6
18. Bluen BE, Lachter J, Khamaysi I, Kamal Y, Malkin L, Keren R, et al,. Accuracy and Quality Assessment of EUS-FNA: A Single-Center Large Cohort of Biopsies. Diag. TherEndosc., 2012: 139563.
19. Lee YN, Moon JH, Kim HK, Choi HJ, Choi MH, Kim DC, et al,. Core biopsy needle versus standard aspiration needle for endoscopic ultrasound-guided sampling of solid pancreatic masses: a randomized parallel-group study. Endosc 2014; 46(12): 1056 – 62

20. Bang JY, Hebert-Magee S, Trevino J, Ramesh J, Varadarajulu S.Randomized trial comparing the 22-gauge aspiration and 22-gauge biopsy needles for EUS-guided sampling of solid pancreatic mass lesions.GastrointestEndosc. 2012; 76(2): 321 –7
21. Sugimoto M, Takagi T, Hikichi T, Suzuki R, Watanabe K, Nakamura J, et al,. Conventional versus contrast enhanced harmonic endoscopic ultrasonography-guided fine-needle aspiration for diagnosis of solid pancreatic lesions. A prospective randomized trial. Pancreatology. 2015; 15(5): 538 –41
22. Rastogi A, Wani S, Gupta N, Singh V, Gaddam S, Reddymaso S, et al,. A prospective, single-blind, randomized, controlled trial of EUS-guided FNA with and without a stylet. GastrointestEndosc. 2011; 74(1): 58-64
23. Mohamadnejad M, Mullady D, Early DS, Collins B, Marshall C, Sams S, et al,. Increasing number of passes beyond 4 does not increase sensitivity of detection of pancreatic malignancy by endoscopic ultrasound-guided fine-needle aspiration.Clin Gastroenterol Hepatol. 2017; 15(7): 1071 –78
24. Chen VK, Eloubeidi MA.EUS guided FNA is superior to lymph node echofeatures: a prospective evaluation of mediastinal and peri intestinal lymphadenopathy: Am J Gastroenterol. 2004:99(4):628-33.
25. Binmoeller KF, Rathod VD. Difficult pancreatic mass FNA: tips for success. GastrointestEndosc. 2002;56:S86-91
26. Polkowski M, Larghi A, Weynand B,Boustière C, Giovannini M, Pujol B, et al,. Learning, techniques, and complications of endoscopic ultrasound (EUS)-guided sampling in gastroenterology: European Society of Gastrointestinal Endoscopy (ESGE) Technical Guideline. Endosc. 2012:44(2);190-206.